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ORIGINAL ARTICLE

High prevalence of *Pneumocystis jirovecii* pneumonia among Mozambican children <5 years of age admitted to hospital with clinical severe pneumonia

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Abstract

We aimed to describe *Pneumocystis jirovecii* pneumonia (PCP) prevalence and features in children from sub-Saharan Africa and to investigate PCP-associated risk factors. During 2006–2007 we used molecular methods to test children younger than 5 years old admitted with severe pneumonia to a hospital in southern Mozambique for *Pneumocystis* infection. We recruited 834 children. PCP prevalence was 6.8% and HIV prevalence was 25.7%. The in-hospital and delayed mortality were significantly higher among children with PCP (20.8% vs. 10.2%, p 0.021, and 11.5% vs. 3.6%, p 0.044, respectively). Clinical features were mostly overlapping between the two groups. Independent risk factors for PCP were age less than a year (odds ratio (OR) 6.34, 95% confidence interval (CI) 1.86–21.65), HIV infection (OR 2.99, 95% CI 1.16–7.70), grunting (OR 2.64, 95% CI 1.04–6.73) and digital clubbing (OR 10.75, 95% CI 1.21–95.56). PCP is a common and life-threatening cause of severe pneumonia in Mozambican children. Mother-to-child HIV transmission prevention should be strengthened. Better diagnostic tools are needed.

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Introduction

Pneumocystis jirovecii pneumonia (PCP) is a common opportunistic infection in immunosuppressed patients, especially those living with HIV. PCP usually affects aggressively the lungs, rapidly causing respiratory failure with marked hypoxemia. In developed countries, PCP remains one of the leading AIDS-

defining diagnoses, both in the United States and Europe, accounting for over 16% of all AIDS diagnoses in Europe in 2008 [1]. PCP incidence has declined since the introduction of highly active antiretroviral therapy (HAART) [2]. Current cases of PCP in adults are associated with undiagnosed HIV cases and with nonadherence to HAART or PCP prophylaxis [3,4]. In children from developed countries, PCP incidence has declined from 25 cases per 1000 HIV-infected children in 1994 to fewer than five cases per 1000 HIV-infected children in 2004 [5]. The most important risk factor for PCP is a CD4 count lower than 200/mm³ in children older than 6 years and adults, or a CD4 proportion lower than 15% in younger children [5–7]. Predictive factors at admission for PCP diagnosis in children include HIV infection, age less than 6 months, high respiratory

rate, low oxygen saturation and absence of fever and vomiting [6,7].

Data on paediatric PCP from developing countries are scarce, especially from sub-Saharan Africa, and are mostly based on small series of cases [8]. The prevalence of *Pneumocystis jirovecii* in African children with severe pneumonia varies from 3.8% to 49% [9–15]. HIV infection and young age were associated with PCP and PCP-associated mortality, which ranged from 20% to 62.5%.

The present study aimed to determine the PCP prevalence among children admitted with severe clinical pneumonia to a district hospital in southern Mozambique and to describe clinical, laboratory, microbiologic and radiologic features. Aetiological investigation among admitted patients permitted a detailed comparison between patients with and without PCP, and the investigation of PCP-associated risk factors.

Methods

Study setting and population

This prospective study was conducted from September 2006 to September 2007 at Manhiça District Hospital, the referral health facility for a rural area in southern Mozambique. The Centro de Investigação em Saúde de Manhiça (CISM) has run a Demographic Surveillance System since 1996, linked with a morbidity surveillance system at Manhiça District Hospital and peripheral health centers [16]. Such surveillance covered, at the time of the study, an area of approximately 500 km² and 80 000 persons. In 2005, the prevalence of HIV among pregnant women attending the antenatal clinic was high (23.6%), and the estimated prevalence among the birth cohort was 3% [17]. At the time of the study, severe pneumonia accounted for 16% of hospitalizations among children younger than 2 years old and had an associated case-fatality rate of 11% [18].

Procedures for recruited children and sample collection

This study was part of a larger project designed to investigate the characteristics of children younger than 5 years old admitted with respiratory distress. Children fulfilling the inclusion criteria and whose parents had signed an informed consent form underwent study procedures. Children presenting with clinical signs for more than 2 weeks and those who were household contacts of known tuberculosis cases were excluded. Anteroposterior chest radiographs were obtained within the first 48 hours of hospitalization. Pulse oximetry (Nellcor, Boulder, CO) was used to determine oxygen saturation, and nasopharyngeal aspirates (NPA) were

collected using NPAK kits (MPRO, Farmington Hills, MI). Venous blood was obtained at admission for malaria diagnosis, blood culture, full blood cell count and biochemical determinations.

X-ray interpretation

Chest X-rays, performed with a Siemens machine, were interpreted blindly by two independent readers following a World Health Organization (WHO)-designed X-ray interpretation protocol [19]. Episodes with consolidation and/or pleural effusion were defined as radiologically confirmed pneumonia (end point pneumonia). Other radiologic end points included interstitial infiltrates or normal radiographs. A third reader interpreted images with discordant results from the two primary readers.

Laboratory methods

Packed cell volume was measured with a haematocrit reader (Hawksley and Sons, Lancing, UK). Thick and thin blood films for malaria diagnosis were processed and examined according to standard methods [16]. Glycaemia was determined using Accu-Chek (Roche, Mannheim, Germany), and blood lactate levels were determined using Lactate Pro (FaCT Canada, Quesnel, British Columbia, Canada) at the bedside. A haematology analyser (Kx21; Sysmex, Denver, CO) and a biochemistry analyser (Vitros DT60; Ortho Clinical Diagnostics, Raritan, NJ) were used. Blood cultures were processed and bacterial isolates identified as previously described [20–22]. The presence of different respiratory viruses (influenza virus A and B, respiratory syncytial virus A and B, parainfluenza virus 1, 2, 3 and 4, adenovirus, human rhinovirus, human metapneumovirus and enterovirus) in NPA was determined using four different PCRs [23].

HIV-specific procedures

Recruited study children were referred for HIV counselling and testing, which required an additional parental consent. HIV-1 serodiagnosis was performed using a sequential testing algorithm with two rapid HIV-1 antibody tests (Determine; Abbott Laboratories, Abbott Park, IL; Unigold; Trinity Biotech, Bray, Ireland). HIV infection was confirmed in seropositive children aged <18 months and older children with discordant cases using HIV-1 DNA Amplicor Test 1.5 (Roche Molecular Systems, Branchburg, NJ). HIV diagnosed children were followed up according to national guidelines.

Molecular diagnosis of *Pneumocystis jirovecii*

Pneumocystis jirovecii infections were investigated in NPA samples using molecular methods. After digestion with proteinase K at 56°C, DNA from *P. jirovecii* was extracted using the

QIAmp DNA commercial Kit (Qiagen, Hilden, Germany). Three specific genes were targeted: (a) nested PCR of mtLSU (mitochondrial large subunit) rRNA locus using pAZ 102-fE/rH (external primer) and pAZ 102-fX/rY (internal primer); (b) nested PCR of mtSSU (mitochondrial small subunit) rRNA locus using pAZ 112-10F/10R (external primer) and pAZ 112-13/14 (internal primer); and (c) touchdown PCR of the DHPS (dihydropteroate synthase) locus using primers DHPS-3 (GCG CCT ACA CAT ATT ATG GCC ATT TTA AAT C) and DHPS-4 (GGA ACT TTC AAC TTG GCA ACC AC) [24,25].

Definitions

Severe pneumonia was defined as cough and/or breathing difficulties, plus increased respiratory rate according to age group and chest indrawing. An increased respiratory rate was defined according to age following standard WHO definitions [26]. Fever was defined as an axillary temperature of $\geq 37.5^{\circ}\text{C}$. Nutritional status was based on weight-for-age z scores, which were calculated using the least mean square method and the 2000 CDC Growth Reference (Centers for Disease Control and Prevention, Atlanta, GA). PCP was defined as having at least two *Pneumocystis jirovecii* genes amplified by PCR in a child with symptoms of pneumonia. This was decided to increase the specificity of the definition. Coagulase-negative staphylococci, *Bacillus* species, or *Micrococcus* species were considered contaminants if isolated in the blood culture.

Case-fatality rates represent in-hospital mortality and do not include patients who were lost from follow-up or were transferred. Mortality occurring within 21 days after hospital discharge (delayed mortality) was investigated with Demographic Surveillance System data for children residing in the CISM study area. Worsening was defined as presence of seizures, deep coma, prostration, low oxygen saturation, dehydration or hypoglycaemia during admission in children without these signs and symptoms at arrival, and as a decrease in oxygen saturation of more than five percentage points at any time of admission compared to saturation at arrival.

The rainy season was defined as November to April and the dry season as May to October.

Case management and treatment

Severe pneumonia was managed according to national guidelines, consistent with the IMCI WHO guidelines [26]. Children requiring specialized care were transferred to Maputo Central Hospital. By the time children were recruited, *Haemophilus influenzae* b or pneumococcal vaccines were not available in Mozambique. National guidelines recommend the use of trimethoprim-sulphamethoxazole for PCP prophylaxis in patients with a confirmed HIV infection.

Data management and statistical analysis

All study questionnaires were double-entered in FoxPro 5.0 (Microsoft, Redmond, WA). Statistical analyses were performed by Stata 13 (StataCorp, College Station, TX).

Categorical variables were compared by chi-square test or Fisher's exact test. Normal distribution was assessed visually. Means of normally distributed variables were compared by Student's t test. For nonnormally distributed variables, medians and interquartile ranges are presented, and the Wilcoxon rank sum test was used to assess differences.

Univariate and multivariate logistic regression analyses were performed to assess associations between explanatory variables and having PCP. For the multivariate analysis, automated backward stepwise estimations were calculated using data from patients with available HIV tests result. All variables associated with PCP infection at a significance level of $p < 0.10$ in the univariate analysis were included in the model. Odds ratios (OR) and 95% confidence intervals (CI) are presented.

Ethical approval

The study was approved by the Mozambican National Bioethics Committee, the Institutional Review Board of the Hospital Clinic (Barcelona, Spain) and the World Health Organization review board.

Results

General overview

During the study period (20 September 2006 to 20 September 2007), 2943 children were admitted to Manhica District Hospital, of whom 926 (31.5%) had severe pneumonia according to the IMCI definition (Fig. 1). The parents of 834 of these patients

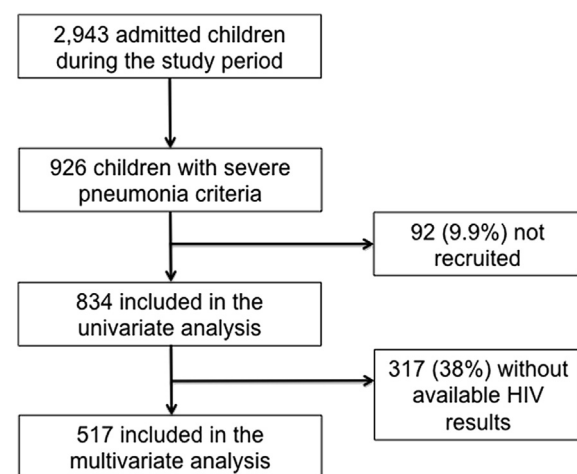


FIG. 1. Study profile.

(28.3% of all admissions; 90.1% of all children who met the study criteria) provided consent that permitted their children to be included in the analysis. Consent for HIV screening was obtained for 517 (62.0%) of the recruited patients, and 133 (25.7%) were HIV positive. The median age was 10.5 months (interquartile range 4.2–20.8), 446 (53.5%) were infants (<12 months old) and 332 (36.8%) were girls. Blood culture results were available for 730 children (87.5%), and 108 of them (14.8%) were positive. The most frequently isolated bacteria were *Streptococcus pneumoniae* (46/108, 42.3% of the positive blood cultures), *Haemophilus influenzae* b (25/108, 23.1%) and enteric Gram-negative bacilli (11/108, 10.2%). Viral detection in NPA was available for 806 children (96.6%), and 392 (48.6%) were positive for respiratory viruses, with multiple infections being common (76/392, 19.4% of positive NPA). Malaria parasitemia results were available for 825 children (98.9%); 106 (12.9%) were positive.

Characteristics of patients according to PCP diagnosis

A total of 77 children (9.2%) had at least one *Pneumocystis jirovecii* gene detected by PCR (Table 1). Among them, 20 (26%) were positive for only one gene, which was in all cases mtLSU rRNA. A total of 57 children (6.8%) had two or more genes identified by PCR, thus fulfilling our PCP definition. Prevalence of PCP among recruited patients was 14.3% (19/133) in HIV-positive children and 3.3% (13/384) in HIV-negative children ($p < 0.001$; OR for being HIV positive among children with PCP, 4.76, 95% CI 2.24–10.09).

Children with a PCP diagnosis were younger than non-PCP children (median age 3.6 months vs. 11.5 months, $p < 0.001$; Table 2), more frequently had gastrointestinal symptoms (diarrhoea and/or vomiting) before admission and had lower oxygen saturation at admission. A total of 47 children were suspected to have PCP on clinical grounds, 19 in the PCP group and 28 in the non-PCP group, yielding a sensitivity of 33.3% and a specificity of 96.4% when compared to our PCR-based case definition.

Regarding laboratory results, abnormal C-reactive protein was more frequently detected in non-PCP children (36.4% vs.

62.4%, $p = 0.017$; Supplementary Table 1). There were no differences between the two groups in the rest of parameters.

Regarding coinfections, HIV prevalence was very high in both groups but was significantly higher in PCP children (59.4% vs. 23.5%, $p < 0.001$). Respiratory viruses were commonly identified in both groups, with human rhinovirus being the most commonly identified virus in both groups (Table 3). Distribution of respiratory viruses was similar in PCP and non-PCP children. Bacteraemia was frequent in both groups (14.6% vs. 14.8%), with *Streptococcus pneumoniae* being the most commonly isolated bacterium in both groups.

Evolution during admission and outcome

Children with PCP experienced a significantly worse disease evolution during admission than non-PCP children (Table 2). The in-hospital case-fatality rate was high in both groups: 20.8% in children with PCP and 10.2% in non-PCP children ($p = 0.021$). Among surviving children from the Demographic Surveillance System area, mortality up to 21 days after discharge (delayed mortality) was also significantly higher in the PCP group (11.5% vs. 3.6%, $p = 0.044$).

HIV infection did not affect the effect of PCP on mortality (Supplementary Table 2; test of interaction $p = 0.459$). On the other hand, PCP seemed to be associated with worsening disease during admission among HIV-positive children, but not among HIV-negative children, although the interaction was not statistically significant (test of interaction $p = 0.213$).

Risk factors independently associated with PCP

In the multivariate analysis (Table 4), the risk of having PCP was independently associated with being an infant, having positive HIV status, having radiologic infiltrates, grunting and digital clubbing. Vomiting, severe malnutrition, unilateral signs at respiratory physical examination and low oxygen saturation at admission were not independently associated with PCP.

Discussion

In our series, *Pneumocystis jirovecii* played an important role in children with severe pneumonia, particularly among HIV-infected children. Overall, HIV prevalence was 25.6% and PCP prevalence 6.8%, which is lower than previously reported in most of the studies conducted in similar settings (3.8–21%) [9,10,12,13,15]. This is probably explained by differences in age, inclusion criteria and HIV prevalence. In previous studies, HIV's prevalence rates paralleled PCP's reported prevalence [12,13]. Specificity of diagnostic tools may also partly explain the differences in PCP prevalence between studies. In the present study, two nested PCR and one touchdown PCR were used,

TABLE 1. *Pneumocystis jirovecii* molecular diagnosis: genes amplified by gene group ($n = 834$ total samples)

Characteristic	mtLSU	mtSSU	DHPS
Only one gene ($n = 20$)	20	0	0
Two genes ($n = 31$)	30	28	4
All three genes ($n = 26$)	26	26	26
Positive samples, n (%)	76 (9.1)	54 (7.3)	30 (3.6)

DHPS, dihydropteroate synthase; mtLSU, mitochondrial large subunit; mtSSU, mitochondrial small subunit.

TABLE 2. Univariate analysis of clinical characteristics, evolution and outcome of patients according to PCP diagnosis

Characteristic	PCP		Non-PCP		p ^a
	Value	n	Value	No.	
Female sex	29 (51)	57	303 (39)	777	0.077
Age (years)	3.6 (2.8–5.0)	57	11.5 (4.8–21.5)	777	<0.001 ^b
Age group		57		777	<0.001
Infants	48 (84.2)		398 (51.2)		
1–5 years	9 (15.8)		379 (48.8)		
HIV-positive status	19 (59.4)	32	114 (23.5)	485	<0.001
Admission during rainy season	33 (57.9)	57	465 (59.9)	777	0.772
Clinical suspicion of PCP	19 (33.3)	57	28 (3.6)	777	<0.001
Clinical history and referred symptoms					
Previous severe malaria	1 (1.8)	57	41 (5.3)	777	0.240
Previous pneumonia admission	3 (5.3)	57	78 (10.0)	777	0.240
Fever	52 (91.2)	57	701 (90.2)	777	0.804
Cough	54 (94.7)	57	705 (90.7)	777	0.308
Breathing difficulty	53 (93.0)	57	679 (87.4)	777	0.213
Diarrhoea	21 (36.8)	57	177 (22.8)	777	0.016
Vomiting	18 (31.6)	57	167 (21.5)	777	0.077
Seizures	1 (1.8)	57	53 (6.8)	777	0.134
Intake refusal	12 (21.1)	57	163 (21.0)	777	0.989
Clinical signs at admission					
Temperature	37.4 (1.0)	57	37.8 (1.3)	777	0.038
Hyperpyrexia ($\geq 39^{\circ}\text{C}$)	7 (12.3)	57	170 (21.9)	777	0.087
Oxygen saturation (%), mean (SD)	83.6 (16.3)	57	91.4 (8.4)	777	<0.001 ^c
Low oxygen saturation (< 90%)	29 (50.9)	57	201 (26.2)	777	<0.001
Tachypnea (age specific)	53 (93.0)	57	686 (88.3)	777	0.282
Nasal flaring	36 (63.2)	57	400 (51.5)	777	0.088
Deep breathing	8 (14.0)	57	77 (9.9)	777	0.320
Crackles	35 (61.4)	57	527 (67.8)	777	0.318
Wheezing	7 (12.3)	57	171 (22.0)	777	0.084
Hypophonesis	1 (1.8)	57	37 (4.8)	777	0.293
Unilateral chest signs	17 (29.8)	57	144 (18.5)	777	0.037
Rhinorrhoea	9 (15.8)	57	109 (14.0)	777	0.713
Grunting	17 (29.8)	57	127 (16.3)	777	0.009
Stridor	0 (0)	57	11 (1.4)	777	0.366
Thoracoabdominal breathing	14 (24.6)	57	75 (9.7)	777	<0.001
Heart gallop	2 (3.5)	57	27 (3.5)	777	0.989
Pallor	7 (12.3)	57	97 (12.5)	777	0.964
Deep coma	0 (0)	57	26 (3.4)	777	0.161
Prostration	12 (21.1)	57	169 (21.8)	777	0.902
Dehydration	8 (14.0)	57	156 (20.1)	777	0.268
Oral candidiasis	10 (17.5)	57	63 (8.1)	777	0.015
Splenomegaly	6 (10.5)	57	81 (10.4)	777	0.981
Digital clubbing	2 (3.5)	57	7 (0.9)	777	0.066
Nutritional status					0.158
WAZ more than –1 SD	16 (28.1)	57	256 (33.3)	768	
WAZ –1 to –3 SD	20 (35.1)	57	318 (41.4)	768	
WAZ less than –3 SD	21 (36.8)	57	194 (25.3)	768	
Worsening during admission	26 (45.6)	57	158 (20.3)	777	<0.001
Oxygen saturation	16 (28.1)		73 (9.4)		<0.001
Prostration	11 (19.3)		64 (8.2)		0.005
Deep coma	0 (0)		21 (2.7)		0.209
Hypoglycaemia	5 (8.8)		11 (1.4)		<0.001
Duration of admission, days. Median (IQR)	6.6 (3.9–11)	57	4.0 (2.7–6.6)	777	<0.001 ^b
In-hospital CFR	10 (20.8)	48	72 (10.2)	709	0.021
Delayed mortality ^d	3 (11.5)	26	17 (3.6)	475	0.044

Data are presented as n (%) or median (IQR) unless otherwise indicated.

CFR, case-fatality rate; IQR, interquartile range; PCP, *Pneumocystis jirovecii* pneumonia; SD, standard deviation; WAZ, weight-for-age z score.

^ap value from chi-square test unless otherwise indicated.

^bp value from Wilcoxon rank sum test.

^cp value from Student t test.

^dMortality during 21 days after discharge; includes only children from the study area.

which have previously shown higher specificity in PCP diagnosis than immunofluorescence [14,27].

The PCP case-fatality rate in our study was 20.8%, similar to the one reported in a large study conducted in South Africa but much lower than others conducted in Uganda, Malawi and South Africa, which ranged 40–63% [9–15]. In some studies patients had a more severe condition or were younger [10,12,13], but not in others [9,15]. The present study was not designed to assess the current management of PCP that could explain these differences. Our reported case-fatality rate associated with PCP remains extremely high.

In our series, clinicians suspected PCP in a third of the cases on the basis of clinical grounds and radiologic results, leaving most PCP cases undiagnosed. Molecular techniques are not routinely available for clinicians in developing countries. Better point-of-care diagnostic methods would therefore be highly desirable, and clinicians should suspect PCP in children who experience a bad response to common antibiotics.

Concurrent isolation of respiratory viruses in NPA and of bacteria in blood culture was common in our study and was more frequent than has been previously reported. In a study in South Africa assessing 231 episodes of severe pneumonia in

TABLE 3. Univariate analysis of microbiology results according to PCP diagnosis

Microbiology result	PCP		Non-PCP		p ^a
	n (%)	n	n (%)	n	
Malaria slide positive	4 (7.1)	56	102 (13.3)	769	0.186 ^b
Positive blood culture	7 (14.6)	48	101 (14.8)	682	0.966 ^b
<i>Streptococcus pneumoniae</i>	4 (8.3)		42 (6.2)		0.540
<i>Haemophilus influenzae</i> b	0 (0)		25 (3.7)		0.395
Enteric Gram-negative bacilli	1 (2.1)		10 (1.5)		0.533
Non-typhi <i>Salmonella</i>	0 (0)		6 (0.9)		1.0
<i>Staphylococcus aureus</i>	1 (2.1)		6 (0.9)		0.385
Other (noncontaminants)	1 (2.1)		11 (1.6)		0.593
Positive CSF	1 (20.0)	5	14 (10.3)	136	0.435
<i>H. influenzae</i> b	0 (0)		9 (6.6)		1.0
<i>S. pneumoniae</i>	1 (20.0)		3 (2.2)		0.147
Other (noncontaminants)	0 (0)		2 (1.5)		1.0
Respiratory virus detected in NPA	29 (50.9)	57	363 (48.5)	749	0.725 ^b
Human rhinovirus	18 (31.6)		176 (23.5)		0.169 ^b
Adenovirus	3 (5.3)		99 (13.2)		0.082 ^b
Respiratory syncytial virus A	2 (3.5)		47 (6.3)		0.569
Human metapneumovirus	4 (7.0)		35 (4.7)		0.348
Parainfluenza virus 1–4	1 (1.8)		30 (4.0)		0.718
Influenza virus A	2 (3.5)		19 (2.5)		0.657
Influenza virus B	3 (5.3)		15 (2.0)		0.108
Enterovirus	2 (3.5)		16 (2.1)		0.368
Virus detection					
Single	23 (79.3)		293 (80.7)		
Double	6 (20.7)		66 (18.2)		
Triple	0 (0)		4 (1.1)		0.811 ^b

CSF, cerebrospinal fluid; NPA, nasopharyngeal aspirate; PCP, *Pneumocystis jirovecii* pneumonia.^ap value from Fisher exact test unless indicated otherwise.^bp value from chi-square test.**TABLE 4.** Multivariate analysis of independent risk factors for PCP

Characteristic	Adjusted OR	95% CI		p
		Lower	Upper	
Age <1 year	6.34	1.86	21.65	0.003
HIV positive	2.99	1.16	7.70	0.023
Grunting	2.64	1.04	6.73	0.042
Digital clubbing	10.75	1.21	95.56	0.033
Radiologic infiltrates	3.56	1.26	10.04	0.017

CI, confidence interval; OR, odds ratio; PCP, *Pneumocystis jirovecii* pneumonia.

HIV-infected children, virus coinfection was found in 11–12% of patients and bacteraemia in 6–10%, compared to 49% and 15% in the present study, respectively [11]. The difference in virus coinfection is probably the result of the diagnostic methods, as multiplex nested PCR was used in our study compared to less sensitive antibody assays used in other studies.

Limitations

The main limitation of this study derives from the lack of a reference standard for PCP diagnosis. According to our definition, the detection of a unique *Pneumocystis* gene on NPA was considered negative, which may have led to misclassifying some true cases. Children with only one positive gene were more similar to children with no positive gene in terms of HIV

prevalence (21% vs. 23%) and proportion of infants (45% vs. 51%) than to children with two to three positive genes (HIV prevalence 59%; proportion of infants 84%). However, the high in-hospital mortality in the subgroup of one positive gene (25%) might be evidence of misclassification of some true cases, which would lead to underestimating differences between the groups considered in the analysis. On the other hand, detection of PCP genes on NPA could be evidence of colonization instead of infection. More invasive samples such as bronchoalveolar lavage or lung biopsy might be useful for differentiating colonization from infection, but the risk of these procedures was considered excessive in symptomatic children.

Another important limitation is that HIV test results were unavailable in 38% of the study children; these data were excluded from the multivariate analysis. We did not collect data on HIV or PCP management, or on immunologic status based on CD4 count or presence of cytomegalovirus in blood (a marker of severe immunosuppression) among HIV-positive children. These are likely to have an effect on PCP-associated mortality and prevalence.

Finally, the effect of smoke exposure, a known risk factor for acquiring community pneumonia [28], was not controlled for. A recent study in the same area showed that 8.7% of the children had at least one parent who smoked, and that less than 2% of the households had their cooking space located inside the house (unpublished data).

Conclusions

PCP was common in the study children admitted with severe pneumonia and was associated with high mortality and clinical worsening of disease during admission. Clinicians should suspect PCP in HIV-positive infants, especially those who experience evolution of disease during admission. However, older children and HIV-negative patients can also have PCP. Clinical signs and laboratory results could not reliably diagnose PCP, but the presence of grunting and digital clubbing should trigger the possibility of its diagnosis. Innovative diagnostic tools need to be urgently developed for those populations most at risk. The performance of the mother-to-child HIV transmission prevention should be investigated and strengthened if needed.

Transparency declaration

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Appendix A. Supplementary data

Supplementary data related to this article can be found online at <http://dx.doi.org/10.1016/j.cmi.2015.07.011>.

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